

On-site test to detect syphilis in pregnancy: a systematic review of test accuracy studies

Rogozińska E^{1,2}, Kara-Newton L¹, Zamora J.R.^{1,3} Khan K.S.^{1,2}

¹Women's Health Research Unit, Blizard Institute, Barts and the London School of Medicine and Dentistry, London, United Kingdom

²Multidisciplinary Evidence Synthesis Hub (mEsh), Centre for Primary Care and Public Health, Blizard Institute, Barts and the London School of Medicine and Dentistry, London, United Kingdom

³Clinical Biostatistics Unit, Hospital Ramon y Cajal (IRYCIS) and CIBER Epidemiology and Public Health, Madrid, Spain

Running title: Accuracy of antenatal tests to detect syphilis

Corresponding author:

Ewelina Rogozińska

Women's Health Research Unit

Barts and The London School of Medicine and Dentistry

Queen Mary University of London

Yvonne Carter Building, E12AB London

Tel: +44 20 7882 5881

Email: e.a.rogozinska@qmul.ac.uk

Other authors:

Lailah Kara-Newton

Women's Health Research Unit

Barts and The London School of Medicine and Dentistry

27 Queen Mary University of London
28 Yvonne Carter Building, E12AB London
29 Email: lailahkn@gmail.com
30
31 Javier R. Zamora
32 Hospital Ramon y Cajal
33 Clinical Biostatistics Unit
34 Crta. Colmenar, km 9.100
35 28034 Madrid
36 Email: javier.zamora@hrc.es
37
38 Khalid S. Khan
39 Women's Health Research Unit
40 Barts and The London School of Medicine and Dentistry
41 Queen Mary University of London
42 Yvonne Carter Building
43 E12AB London
44 Email: k.s.khan@qmul.ac.uk
45 Tel: +44 20 7882 2525
46

47 **Abstract**

48 **Background** Syphilis in pregnancy can lead to fetal and neonatal death or congenital
49 anomalies. Accurate on-site tests are an essential part of effective prevention of mother-to-
50 child transmission of the disease.

51 **Objective** This systematic review assessed the accuracy of the on-site tests to detect infection
52 with *Treponema pallidum* in pregnant women.

53 **Search strategy** Major databases were searched from inception to January 2016 using terms:
54 “pregnancy”, “antenatal”, “syphilis”, “*Treponema pallidum*” with their variations, and the
55 search limit for the relevant study design.

56 **Selection criteria** We included studies that used dual reference standard (non-treponemal and
57 treponemal tests) to detected syphilis in pregnancy.

58 **Data collection and analysis** Extracted accuracy data were tabulated and pooled using
59 hierarchical, bivariate random effects model.

60 **Main results** Seven studies (combined sample 17,546) reporting the accuracy of four on-site
61 tests met the eligibility criteria. On average, Determine™ and SD BioLine Syphilis 3.0 had
62 the highest sensitivity out of all evaluated tests 0.83 (95% CI 0.58, 0.98) and 0.86 (95% CI
63 0.82, 0.89), respectively with a high specificity 0.96 (95% CI 0.89, 1.00) and 0.99 (95% CI
64 0.94, 1.00), respectively. Qualitative Rapid Plasma Reagin card commonly used in clinical
65 practice had a pooled sensitivity of 0.70 (95% CI 0.54, 0.88) and specificity of 0.97 (95% CI
66 0.96, 0.99).

67 **Conclusion** Immunochromatographic tests such as Determine and SD BioLine Syphilis 3.0
68 seem to be acceptable options in antenatal testing for syphilis, especially in resource-limited
69 settings. Future research should seek more evidence to strengthen this claim.

70 **Keywords** Syphilis, Antenatal care, Test accuracy, On-site test

71 **Tweetable abstract** On-site test to detect syphilis - options during antenatal care

72 **Introduction**

73 Syphilis, a sexually transmitted infection caused by the bacterium *Treponema pallidum*
74 (*T.pallidum*), is endemic throughout the developing world.(1) Infection until one year is
75 classified as early syphilis, and after one year as late syphilis. The initial manifestation of the
76 disease can be easily overlooked and progress to the secondary stage which if undiagnosed
77 and consequently non-treated leads to a period of latency with no visible signs of the disease.
78 The infection is most commonly transmitted through sexual intercourse, and it can also be
79 passed from mother to a child; in utero or during birth.

80

81 Transmission of the infection had been linked with the birth of children with reactive
82 serology, long-term congenital abnormalities, miscarriages, and fetal and neonatal deaths.
83 (1,2) The World Health Organization (WHO) estimated that in 2008 around 1.36 million
84 pregnant women were expected to have an active form of syphilis. Without any screening or
85 treatment in place these women would have experienced, overall, more than 700,000 adverse
86 outcomes where more than half would be fetal or neonatal deaths.(3)

87

88 In order to prevent mother-to-child transmission of syphilis WHO advocates screening of all
89 pregnant women antenatally and treating those identified with the disease and their
90 partners.(4) The ideal Point-Of-Care (POC) test should be affordable, sensitive, specific, user-
91 friendly, rapid and robust, equipment free, and deliverable to those who need them.

92 Development of POC test has made syphilis testing more accessible especially in low-
93 resource settings, as lengthy and skilled laboratory testing can be avoided.(5)

94 Immunochromatographic tests or the on-site Rapid Plasma Reagin cards performed on-site
95 give healthcare professionals an opportunity to administer treatment immediately and prevent
96 the transmission of the disease.(6)

97

According to reviews assessing the accuracy of the immunochromatographic POC treponemal tests (7,8) they offer an alternative to laboratory-based diagnosis in resource-limited settings. However, none of the reviews focuses solely on pregnant women or compare the immunochromatographic with commonly used in clinics qualitative Rapid Plasma Reagin card which is not an ideal gold standard.(9) Our focus was to synthesise the accuracy of on-site tests used in antenatal care settings to detect syphilis using an established algorithm as a reference standard.(10)

Methods

We conducted the review and reported our findings in compliance with the current guidelines.(11) We searched Medline, Embase, Web of Science, Scopus, and Lilacs with no language restrictions. The original search run from inception to February 2015 was updated in January 2016 (Figure 1). The literature search strategy combined clinical terms such as ‘Pregnancy’, ‘Antenatal’, ‘Gestation’, ‘Treponema pallidum’ and ‘Syphilis’ with a filter for test accuracy studies.(12) The detailed search strategy is available in Appendix S1.

Study selection

Two independent reviewers (ER and LKN) screened references and then full text of potentially relevant articles. The study had to meet following eligibility criteria: recruit pregnant women without symptoms of syphilis (chancre, rash); use as a double reference standard comprising of non-treponemal (the Rapid Plasma Reagin test or venereal disease research laboratory (VDRL)) followed by treponemal test (treponema pallidum haemagglutination assay (TPHA), fluorescent treponemal antibody-absorbed (FTA-Abs) or the treponema pallidum particle agglutination (TPPA) test). Diagnosis of recently contracted infection with *T.palladium* was defined as a positive result on both treponemal and non-treponemal test.(13)

124

125 We excluded studies in which the population showed symptoms of syphilis, women in labour
126 and studies where reference standard was only a treponemal or non-treponemal test. We
127 excluded studies with a case-control design and those where it was not possible to calculate
128 True Positives, False Positives, False Negatives and True negatives. At each stage of the
129 review process, the consensus was reached through a discussion. In the case of a stalemate,
130 the opinion of a third reviewer's was sought (KSK). We did not attempt to contact the study
131 authors for any further information.

132

133 *Data extraction and study quality assessment*

134 All relevant data from included studies were extracted to a standardized, and pre-piloted form.
135 Information about the country, settings, women's characteristics, type of index test and
136 reference standard, and type of collected blood sample were extracted and tabulated. We
137 classified the countries where the studies were conducted by their income following the
138 World Bank ranking.(14)

139

140 The quality of each included study was assessed by two review authors (ER, LKN) using the
141 QUADAS-2 tool.(15) The risk of bias was evaluated for participants' selection, use and
142 interpretation of index test and reference standard, and participants flow and timing. First
143 three aspects were also evaluated in the context of applicability to the review question. The
144 review authors classified each item as "low" (sufficiently addressed), "high" (insufficiently
145 addressed), or "unclear" (insufficient detail presented to allow judgment to be made) risk of
146 bias. We considered a study to be of low risk of bias if; the patients were selected
147 consecutively or randomly, the index and reference standard tests were correctly implemented,
148 and all patients received the reference standard tests.

149

Data synthesis

To construct two-by-two tables we extracted true positive, false positive, true negative, and false negative results or recalculated the numbers from available parameters (sensitivity, specificity, positive predictive value and negative predictive value). All analyses were performed using STATA version 12.1 (College Station, TX: StataCorp LP). Sensitivity, specificity, likelihood ratios for positive and negative test result and 95% confidence intervals (CIs) were computed for all individual studies. Where we had a sufficient number of studies (more than four), we pooled the accuracy parameters using hierarchical, bivariate, random effects model using the multilevel mixed logistic regression model as implemented by *metandi* command.(16) For meta-analysis with less than four studies, we pooled accuracy of sensitivity and specificity, and likelihood ratios separately using *metaprop* and *metan* commands, respectively. Between-study heterogeneity of studies was assessed graphically evaluating forest plots for sensitivity and specificity. Publication bias was not assessed due to lack of consensus over the reliability of currently available methods.(17,18)

Results

The database searches retrieved 2,045 relevant citations; additional eight records were identified through the reference check. Out of 59 potentially relevant articles evaluated by their full text, seven publications met the eligibility criteria (Figure 1). A detailed list of excluded studies with reasons for their exclusion can be found in Table S1.

Characteristics of included studies

Eligible studies recruited combined number of 17,546 pregnant women. The prospective studies were published between 1993 and 2015, with seroprevalence of syphilis ranging from 1 - 11%. In three publications authors didn't mention in the text whether women were previously treated for syphilis,(19-21) one excluded this group (22), and in the remaining

studies around 7% of participants were previously diagnosed with syphilis.(23-25) Included publications reported accuracy data of three immunochromatographic tests: Determine™ (Abbott Laboratories, Chicago, USA), SD BioLine Syphilis 3.0 (Standard Diagnostics Inc., Republic of Korea), VisiTest Syphilis (Omega Diagnostics, Alloa, Scotland) and the qualitative Rapid Plasma Reagin card (multiple manufacturers). The majority of studies recruited women in hospital settings,(19,20,22,23,25) one in primary care (24) and one in the general health centre (21). Three studies were conducted in upper-middle income countries, two in lower-middle income countries and two studies were in low-income countries (Table 1). All studies used fresh blood samples.

Quality assessment

Six out of seven studies had an unclear risk of bias for the sample selection due to a lack of information about the selection process. The majority of studies were assessed as low risk of bias for the implementation of the reference standard and all for the index test. The bias for flow and timing was unclear in two studies due insufficient level of information (Table 2). One study (25) was classified as of high concern over applicability in sample selection as it reports physical examination findings of participants (Table 2). There was no overall concern applicability of included studies in terms of index test and applied reference standard.

Accuracy of immunochromatographic tests

Two studies (20,24) with a combined sample size of 9,587 women reported accuracy data of the Determine™ test. Pooled sensitivity and specificity of the Determine™ were 0.83 (95% CI 0.58, 0.98) and 0.96 (95% CI 0.89, 1.00), respectively with likelihood ratio for the positive test of 24.88 (95% CI 4.19, 147.57), and for a negative test result of 0.16 (95% CI 0.04, 0.66). Two studies (22,25) reported the data on the accuracy of the SD BioLine Syphilis 3.0. Pooled sensitivity from those studies was of 0.86 (95% CI 0.82, 0.89), and sensitivity of 0.99 (95%

CI 0.94, 1.00). The likelihood ratio for the positive and negative test result was 54.87 (95% CI 6.52, 461.65) and 0.15 (95% CI 0.12, 0.20), respectively. The accuracy of the third test, VisiTest Syphilis, was reported in one study of 712 women. (23) The sensitivity of VisiTest was 0.63 (95% CI 0.31, 0.86) and specificity 0.98 (95% CI 0.97, 0.99).

Qualitative Rapid Plasma Reagin card

The qualitative Rapid Plasma Reagin test was used as an index test in five studies. (19-21,23,25) Pooled sensitivity was 0.70 (95% CI 0.50, 0.84) and pooled specificity 0.97 (95% CI 0.96, 0.98). The derived likelihood ratio of the positive test result was 27.07 (95% CI 15.39, 47.61) and the negative result of 0.31 (95%CI 0.17, 0.56). There was visible greater heterogeneity between sensitivity estimates than specificity with the 95% predictive region covering less than one-third of the operating space (Figure S1). The accuracy parameters of all evaluated tests have been collated and summarised in Table 3. The numbers used to calculate the parameters are available in Table S2.

Discussion

Main findings

SD BioLine Syphilis 3.0 test had, on average, the highest sensitivity out of all evaluated immunochromatographic tests, and visibly higher sensitivity than qualitative Rapid Plasma Reagin card. Specificity did not differ significantly between the identified tests.

Strengths and limitations

This systematic review was conducted using following current methodological standards.(11) The use of search limit for test accuracy studies (12), was a pragmatic choice. The search without the limit had too-broad approach to be practicable. Even though, we identified the majority of studies with antenatal population included in the previous reviews and two

additional ones (19,22) the overall number of studies available for the analyses was small. The bivariate analysis was possible only for the RPR card, yet its findings are weakened by a visible heterogeneity of sensitivity parameters between the individual studies.

Test accuracy studies are prone to numerous sources of bias due to patients' selection and retention in the study, implementation of the index test and reference standard. In our review, we managed to limit spectrum bias by excluding studies with case-control design. However, the majority of included studies failed to describe recruitment method and inclusion criteria.

The risk of bias and concern over the applicability of the index tests and reference standards were generally low. Ideally, the reference standard and the index test should be entirely independent of each other.(26) This was true for the immunochromatographic test, yet the lab-based confirmatory algorithm for the qualitative Rapid Plasma Reagin card had as its non-treponemal component quantitative Rapid Plasma Reagin test. This raises concern over an incorporation bias (26), however, the extent to which use of the Rapid Plasma Reagin test as a part of gold standard could distort the results is unclear, and couldn't be avoided due to studies' design.

The average prevalence of double reactive sera in studies evaluating the accuracy of Determine™, SD BioLine Syphilis 3.0, VisiTest Syphilis and the qualitative Rapid Plasma Reagin card were 4.0%, 8.2%, 1.1% and 5.7%, respectively. This level of prevalence is higher than the global prevalence of the disease among antenatal care attendee and in some cases (South Africa or Senegal) even significantly higher than in the countries where the studies were conducted.(27) By definition, sensitivity and specificity do not depend on the disease prevalence. However, their parallel variability can occur due to clinical or artefactual mechanisms.(28) Clinicians before drawing any conclusion basing on the accuracy findings

should be very clear about the clinical question they want to address. The diversity of the prevalence, statistical methods used to pool the data and the quality of reporting impacts the generalisability of presented findings.

The timely delivery of treatment during prenatal period alters the risk of adverse outcomes due to syphilis infection. (29) In order to optimise the applicability of our findings to the context of antenatal care, we defined a clear research question. We focused solely on pregnant women during the perinatal period. We looked for the immunochromatographic, in detecting double positive sera to non-treponemal and treponemal components of the reference standard.

Interpretation

Two previous reviews address the issue of accuracy of the rapid, on-site testing using different methods of data synthesis.(7,8) The first review found that the immunochromatographic tests have a high sensitivity and higher specificity comparable with parameters of non-treponemal.(8) In systematic review with Bayesian approach to data synthesis the Determine test had the highest sensitivity when comparing with *T.palladium* specific reference standard. However, the authors admitted in their work that due to applied methodology the values of sensitivity were overestimated.(7) Both reviews included women tested in antenatal care settings, including women in labour, and focusing on the accuracy and value of the immunochromatographic test in rapid testing for syphilis.

Similar to the previous reviews (7, 8), the immunochromatographic tests were characterised by high sensitivity and specificity. Additionally, their average sensitivity was higher than for the qualitative Rapid Plasma Reagin on-site card (except VisiTech Syphilis) with the average specificity comparable between all the tests. The immunochromatographic tests are comparable in cost (8) and easier to operate than Rapid Plasma Reagin card (21,24) what

makes them less prone to an operator error. The average cost in low resource settings is U.S. \$0.91 and U.S. \$1.05 for the RPR and ICS tests. (8) Nonetheless, their reliability depends on the background proportion of women with past-treated infection who may still test as positive, and consequently be treated unnecessarily. Furthermore, the tests can also give a positive result in various non-venereal treponematoses such as yaws and pinta, these would be considered false positive results and are preferred to false negative results and there is greater benefit in over-treating all patients with positive results as opposed to the alternative.

In the high-prevalence settings (assumed 11%) around 9% of all positive tests with SD BioLine Syphilis 3.0 would be falsely positive in contrast to 21 – 28% with the other immunochromatographic tests or the Rapid Plasma Reagin card. The proportion of potentially missed cases would be 2% for SD BioLine Syphilis 3.0 and Determine™, and 4% for VisiTech and Rapid Plasma Reagin card. Syphilis in pregnancy is effectively treated with penicillin with benzathine penicillin remaining the first-line therapy for early syphilis. (30) The treatment is administered by intramuscular injection and requires three large doses once weekly for three weeks. This requires patients to return to health care services for each dose which may prove difficult in rural settings. With no cases of antibiotic resistance reported so far (31) prevention of mother-to-child transmission of the disease is more important than overtreatment.

Conclusion

Our systematic review adds to the current body of evidence on the accuracy of the rapid and Point-of-Care test to detect infection with *T.palladium* in the context of the antenatal care. Future test accuracy studies should aim to improve reporting of their findings and directly compare the accuracy of available tests controlling for the confounders.

When testing antenatally for syphilis immunochromatographic tests such as Determine™ and SD BioLine Syphilis 3.0 seem to be acceptable options. However, future research is needed to provide more evidence to strengthen this claim.

Acknowledgements

The authors would like to acknowledge the assistance of the following advisors from the WHO Department of Reproductive Health and Research (A. Metin Gülmezoglu, Özge Tunçalp, and Teodora Wi).

Contribution to Authorship

ER selected eligible texts, data extraction form, extracted data, wrote the protocol, cleaned and analysed the data, drafted and revised the manuscript. LKN selected eligible texts, extracted data, and drafted and revised the manuscript. JZ supervised statistical analysis and revised the manuscript. KSK resolved discrepancies between reviewers and revised the manuscript.

Declaration of interest

The authors report no conflict of interest. The ICMJE disclosure forms are available as online supporting information.

Details of ethics approval

Ethical approval was not required for this project.

Funding

This work was conducted as a part of the work stream for the WHO recommendations on antenatal care.

Reference List

- (1) Cohen SE, Klausner JD, Engelman J, Philip S. Syphilis in the modern era: an update for physicians. *Infect Dis Clin North Am* 2013 Dec;27(4):705-22.
- (2) Gomez GB, Kamb ML, Newman LM, Mark J, Broutet N, Hawkes SJ. Untreated maternal syphilis and adverse outcomes of pregnancy: a systematic review and meta-analysis. *Bull World Health Organ* 2013 Mar 1;91(3):217-26.
- (3) Newman L, Kamb M, Hawkes S, Gomez G, Say L, Seuc A, et al. Global estimates of syphilis in pregnancy and associated adverse outcomes: analysis of multinational antenatal surveillance data. *PLoS Med* 2013;10(2):e1001396.
- (4) WHO. The Global Elimination of Congenital Syphilis: rationale and strategy for action. 2007. apps.who.int/iris/bitstream/10665/43782/1/9789241595858_eng.pdf
- (5) Peeling RW, Ye H. Diagnostic tools for preventing and managing maternal and congenital syphilis: an overview. *Bull World Health Organ* 2004 Jun;82(6):439-46.
- (6) WHO The Sexually Transmitted Diseases Diagnostics Initiative (SDI). The use of rapid syphilis tests. 2006 WHO reference number: WHO/TDR/SDI/06.1 www.who.int/reproductivehealth/publications/rtis/TDR_SDI_06_1/en/
- (7) Jafari Y, Peeling RW, Shivkumar S, Claessens C, Joseph L, Pai NP. Are *Treponema pallidum* specific rapid and point-of-care tests for syphilis accurate enough for screening in resource limited settings? Evidence from a meta-analysis. *PLoS One* 2013;8(2):e54695.
- (8) Tucker JD, Bu J, Brown LB, Yin YP, Chen XS, Cohen MS. Accelerating worldwide syphilis screening through rapid testing: a systematic review. *Lancet Infect Dis* 2010 Jun;10(6):381-6.
- (9) Centers for Disease Control. Guidelines for the Prevention and Control of Congenital Syphilis. *MMWR* 37 (suppl no S-1). 1988.
- (10) Morshed MG, Singh AE. Recent trends in the serologic diagnosis of syphilis. *Clin Vaccine Immunol* 2015 Feb;22(2):137-47.
- (11) Leeflang MM, Deeks JJ, Gatsonis C, Bossuyt PM. Systematic reviews of diagnostic test accuracy. *Ann Intern Med* 2008 Dec 16;149(12):889-97.
- (12) Vincent S, Greenley S, Beaven O. Clinical Evidence diagnosis: Developing a sensitive search strategy to retrieve diagnostic studies on deep vein thrombosis: a pragmatic approach. *Health Info Libr J* 2003 Sep;20(3):150-9.
- (13) Ratnam S. The laboratory diagnosis of syphilis. *Can J Infect Dis Med Microbiol* 2005 Jan;16(1):45-51.
- (14) The World Bank Group. Country and Lending Groups. data.worldbank.org/about/country-and-lending-groups [Accessed 5-2-2016]

- 372 (15) Whiting PF, Rutjes AW, Westwood ME, Mallett S, Deeks JJ, Reitsma JB, et al.
373 QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies.
374 Ann Intern Med 2011 Oct 18;155(8):529-36.
- 375 (16) Harbord RM, Whiting P. metandi: Meta-analysis of diagnostic accuracy using
376 hierarchical logistic regression. The Stata Journal 2009;9(2):211-29.
- 377 (17) Deeks JJ, Macaskill P, Irwig L. The performance of tests of publication bias and other
378 sample size effects in systematic reviews of diagnostic test accuracy was assessed. J
379 Clin Epidemiol 2005 Sep;58(9):882-93.
- 380 (18) Song F, Khan KS, Dinnes J, Sutton AJ. Asymmetric funnel plots and publication bias
381 in meta-analyses of diagnostic accuracy. Int J Epidemiol 2002 Feb;31(1):88-95.
- 382 (19) Delport SD. On-site screening for maternal syphilis in an antenatal clinic. S Afr Med J
383 1993 Oct;83(10):723-4.
- 384 (20) Tinajeros F, Grossman D, Richmond K, Steele M, Garcia SG, Zegarra L, et al.
385 Diagnostic accuracy of a point-of-care syphilis test when used among pregnant
386 women in Bolivia. Sex Transm Infect 2006 Dec;82 Suppl 5:v17-v21.
- 387 (21) Van DE, Van d, V, Ndoye I, Piot P, Meheus A. Evaluation of the rapid plasma reagin
388 "teardrop" card test for screening of syphilis in field conditions. Sex Transm Dis 1993
389 Jul;20(4):194-7.
- 390 (22) Kashyap B, Sagar T, Kaur IR. Utility of immunochromatographic assay as a rapid
391 point of care test for screening of antenatal syphilis. Indian J Sex Transm Dis 2015
392 Jul;36(2):162-5.
- 393 (23) Benzaken AS, Sabido M, Galban E, Pedroza V, Araujo AJ, Peeling RW, et al. Field
394 performance of a rapid point-of-care diagnostic test for antenatal syphilis screening in
395 the Amazon region, Brazil. Int J STD AIDS 2011 Jan;22(1):15-8.
- 396 (24) Bronzan RN, Mwesigwa-Kayongo DC, Narkunas D, Schmid GP, Neilsen GA, Ballard
397 RC, et al. On-site rapid antenatal syphilis screening with an immunochromatographic
398 strip improves case detection and treatment in rural South African clinics. Sex Transm
399 Dis 2007 Jul;34(7 Suppl):S55-S60.
- 400 (25) Montoya PJ, Lukehart SA, Brentlinger PE, Blanco AJ, Floriano F, Sairosse J, et al.
401 Comparison of the diagnostic accuracy of a rapid immunochromatographic test and
402 the rapid plasma reagin test for antenatal syphilis screening in Mozambique. Bull
403 World Health Organ 2006 Feb;84(2):97-104.
- 404 (26) Schmidt RL, Factor RE. Understanding sources of bias in diagnostic accuracy studies.
405 Arch Pathol Lab Med 2013 Apr;137(4):558-65.
- 406 (27) WHO. Antenatal care attendees who were positive for syphilis - data by country.
407 apps.who.int/gho/data/node.main.A1359STI [Accessed 5-2-2016]
408
- 409 (28) Leeflang MM, Bossuyt PM, Irwig L. Diagnostic test accuracy may vary with
410 prevalence: implications for evidence-based diagnosis. J Clin Epidemiol 2009
411 Jan;62(1):5-12.

- 412 (29) Hawkes SJ, Gomez GB, Broutet N. Early antenatal care: does it make a difference to
413 outcomes of pregnancy associated with syphilis? A systematic review and meta-
414 analysis. PLoS One 2013;8(2):e56713.
- 415 (30) Walker GJ. Antibiotics for syphilis diagnosed during pregnancy. Cochrane Database
416 Syst Rev 2001;(3):CD001143.
- 417 (31) Genc M, Ledger WJ. Syphilis in pregnancy. Sex Transm Infect 2000 Apr;76(2):73-9.
418
419

420 **Legends**

421 **Figure 1** Study selection diagram

422 **Table 1** Characteristics of studies of on-site tests to detect syphilis among pregnant women

423 **Table 2** Quality assessment of included studies using QUADAS-2 tool

424 **Table 3** Accuracy of tests to detect syphilis among pregnant women

425

426 **Supporting Information**

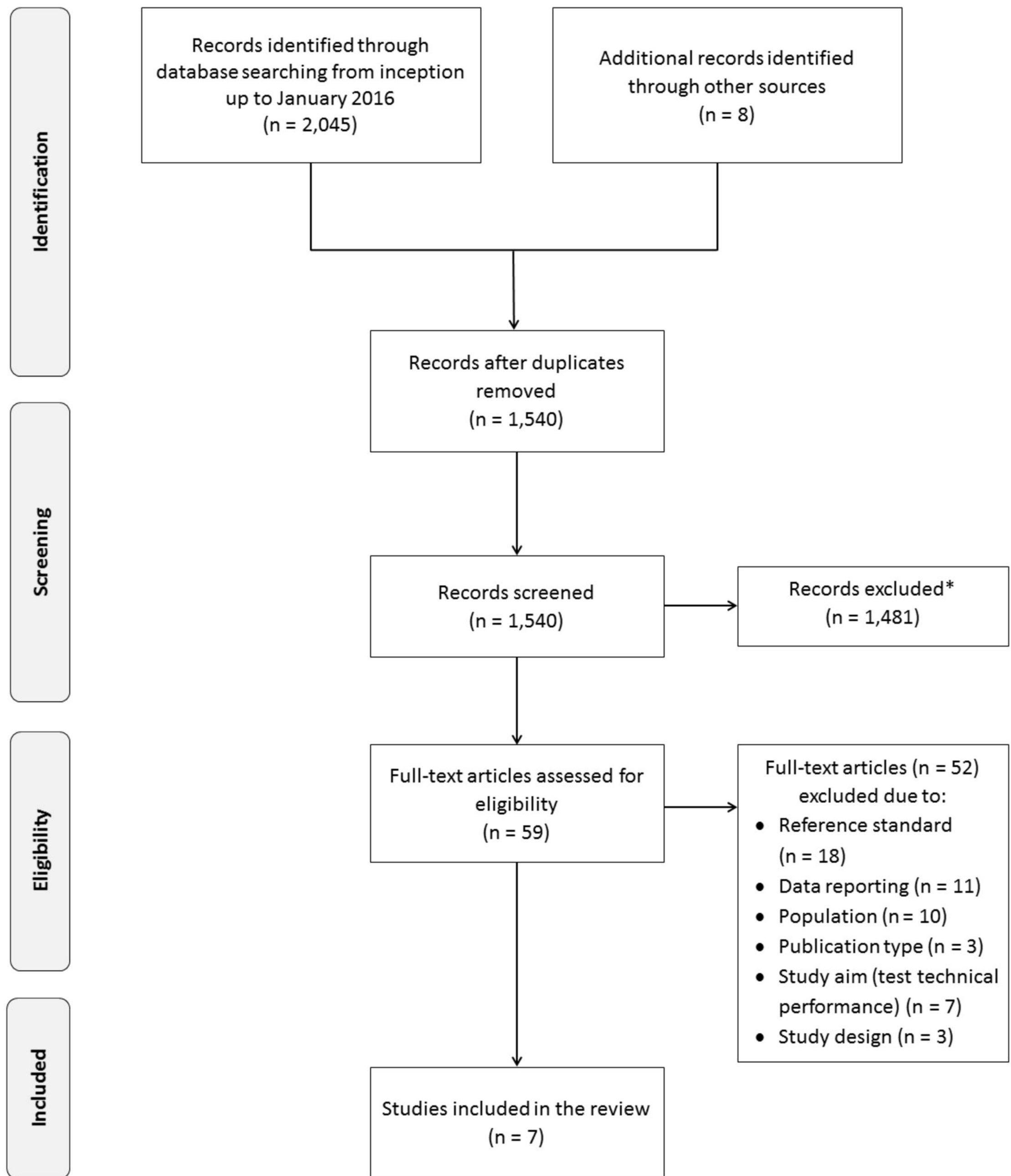
427 **Figure S1** Summary Point in Receiver Operating space for qualitative Rapid Plasma Reagin
428 card

429 **Appendix S1** Search Strategy for Medline 15th January 2015 (updated 11th January 2016)

430 **Table S1** List of excluded full text articles with reasons for exclusion

431 **Table S2** Test accuracy data extracted from included studies

432



**full text of nine papers was not available for the assessment*

Table 1 Characteristics of studies of on-site tests to detect syphilis among pregnant women

Study ID	Country	Settings	Sample size	Reference standard		Type of the index test	Index test	Type of blood sample	Sero-prevalence* (95% CI)
Benzaken 2011	Brazil	Antenatal clinic	712	VDRL	FTA-Abs	Treponemal test - ICS	VisiTest Syphilis test	Whole blood	0.01 (0.01, 0.02)
Bronzan 2007	South Africa	Primary Care clinic	1,250	Quantitative RPR	TPHA	Treponemal test - ICS	Determine™	Whole blood	0.06 (0.05, 0.08)
						Non-treponemal test - RPR	Qualitative RPR card	Whole blood	
Delport 1993	South Africa	Antenatal clinic	1,237	Quantitative RPR	TPHA	Non-treponemal test -RPR	Qualitative RPR card	Plasma	0.07 (0.05, 0.08)
Kashyap 2015	India	University Hospital	200	VDLR	TPHA	Treponemal test - ICS	SD BioLine Syphilis	Serum	0.02 (0.01, 0.05)
Montoya 2006	Mozambique	Antenatal clinic	4,789	Quantitative RPR	TPHA	Treponemal test - ICS	SD BioLine Syphilis	Whole blood	0.08 (0.08, 0.09)
						Non-treponemal test - RPR	Qualitative RPR card	Whole blood	
Tinajeros 2006	Bolivia	Maternity Hospital	8,892	Qualitative RPR	TPPA	Treponemal test - ICS	Determine™	Whole blood	0.04 (0.03, 0.04)
						Non-treponemal test - RPR	Qualitative RPR card	Serum	
Van Dyck 1993	Senegal	Health Centre	466	Quantitative RPR	TPHA/FTA-Abs**	Non-treponemal test - RPR	Qualitative RPR card	Whole blood	0.11 (0.08, 0.14)

*reactive both non-treponemal and treponemal tests; ** on discordant samples

RPR - Rapid Plasma Reagin

ICS - Immunochromatographic strip

FTA-Abs - Fluorescent treponemal antibody absorption

TPHA - Treponema pallidum hemagglutination assay

TPPA - Treponema pallidum particle agglutination assay

VDRL - Venereal disease research laboratory

Table 2 Quality assessment of included studies using QUADAS-2 tool

QUADAS	Risk of bias				Concern over applicability		
Study ID	Sample selection	Index test	Reference standard	Flow and timing	Sample selection	Index test	Reference standard
Benzaken 2011	Low	Low	Low	Low	Unclear	Low	Low
Bronzan 2007	Unclear	Low	Low	Low	Unclear	Low	Low
Delpont 1993	Unclear	Low	Low	Unclear	Unclear	Low	Low
Kashyap 2015	Unclear	Low	Unclear	Low	Low	Low	Low
Montoya 2006	Unclear	Low	Low	Low	High	Low	Low
Tinajeros 2006	Unclear	Low	Low	Unclear	Unclear	Low	Low
Van Dyck 1993	Unclear	Low	Low	Low	Unclear	Low	Low

Table 3 Accuracy of tests to detect syphilis among pregnant women

Index test	Study ID	Reactive/ Non-reactive	Sensitivity (95%CI)	Specificity (95%CI)	Likelihood ratio for a positive test result (95%CI)	Likelihood ratio for a negative test result (95%CI)
Determine	Tinajeros 2006	342/8,850	0.92 (0.88, 0.95)	0.99 (0.98, 0.99)	61.33 (51.49, 73.04)	0.08 (0.06, 0.12)
	Bronzan 2007 [^]	44/651	0.70 (0.56, 0.82)	0.93 (0.91, 0.95)	9.97 (7.11, 13.98)	0.32 (0.20, 0.50)
	Pooled estimates	386/9,201	0.83 (0.58, 0.98)	0.96 (0.89, 1.00)	24.88 (4.19, 147.57)	0.16 (0.04, 0.66)
SD BioLine Syphilis 3.0	Montoya 2006	381/4,105	0.86 (0.82, 0.89)	0.97 (0.96, 0.97)	26.41 (22.23, 31.37)	0.15 (0.12, 0.19)
	Kashyap 2015	4/196	0.75 (0.30, 0.95)	1.00 (0.98, 1.00)	275.80 (16.32, 4660.18)	0.30 (0.08, 1.15)
	Pooled estimates	385/4,301	0.86 (0.82, 0.89)	0.99 (0.94, 1.00)	54.87 (6.52, 461.65)	0.15 (0.12, 0.20)
VisiTech Syphilis	Benzaken 2011 ^{^^}	8/704	0.63 (0.31, 0.86)	0.98 (0.97, 0.99)	40.00 (18.07, 88.57)	0.38 (0.16, 0.93)
Qualitative Rapid Plasma Reagin card	Bronzan 2007 [^]	35/520	0.46 (0.29, 0.63)	0.97 (0.95, 0.98)	14.86 (8.13, 27.14)	0.56 (0.41, 0.76)
	Van Dyck 1993	50/402	0.46 (0.32, 0.61)	0.97 (0.94, 0.98)	13.21 (7.28, 23.97)	0.56 (0.43, 0.72)
	Montoya 2006	381/4,105	0.71 (0.67, 0.76)	0.96 (0.96, 0.97)	19.80 (16.70, 23.48)	0.30 (0.25, 0.35)
	Tinajeros 2006	342/8,847	0.76 (0.71, 0.80)	0.99 (0.99, 0.99)	82.98 (66.01, 104.33)	0.25 (0.20, 0.30)
	Delpont 1993	83/1,154	0.93 (0.85, 0.97)	0.96 (0.95, 0.97)	24.90 (18.46, 33.59)	0.75 (0.04, 0.16)
	Pooled estimates	891/14,728	0.70 (0.50, 0.84)	0.97 (0.96, 0.98)	27.07 (15.39, 47.61)	0.31 (0.17, 0.56)

[^] combined high & low titre (both define active syphilis)^{^^} Missing VDRL samples assumed as positive